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Defining a termite sampling protocol for ecological studies: An effective method to increase statistical power

Cristian S. Dambros[a,b,*], José Wellington Morais[b], Alexandre Vasconcellos[c], Elizabeth Franklin[b]

[a] Universidade Federal de Santa Maria, Centro de Ciências Naturais e Exatas, Departamento de Ecologia e Evolução, Santa, Maria, 97105-900, RS, Brazil
[b] Coordenacão de Biodiversidade, Instituto Nacional de Pesquisas da Amazônia, CP 2223, 69067-375, Manaus, AM, Brazil
[c] Departamento de Sistemática e Ecologia, Laboratório de Termitologia, Universidade Federal da Paraíba, CCEN, 58051-900, João Pessoa, PB, Brazil

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ABSTRACT

Protocols for sampling soil fauna are usually designed to increase the number of species found when one or a few plots are heavily sampled. However, no previous study has evaluated how the number of plots sampled affect the power of statistical tests associating community composition and the environment. We test the effect of the number of transects (plots) and the sampling effort in each transect on the representation of trophic groups and on the association of termite species composition with environmental variables. Data were collected in 30 250m-long transects located in central Amazonia. Each transect was subdivided into 10 non-contiguous sections of 5 m × 2 m each (separation of 22 m between sections). We rarefied the data to determine arrangement of transects and sections that 1) best characterizes the distribution of species in trophic groups and 2) maximizes the chances to detect true associations between termite species composition and environmental predictor variables. When more than six transects (plots) were sampled with at least five non-contiguous sections each, the distribution of trophic groups was similar to the known distribution for the area. However, the detection of the association between termite species composition and environmental variables was more easily detected by increasing the number of transects sampled (plots) than the sampling effort per transect. Our results suggest that spreading sampling effort into at least 15 transects (plots) improves the ability to detect trophic groups and the performance of regression tests associating the composition of species with the environment.

1. Introduction

Termite sampling can be costly and labor intensive, usually requiring active searching and digging [1–4]. The limitations for sampling termites and the necessity to standardize studies have led to the development of protocols that maximize the collection of multiple species in few transects [2]. Statistical analyses commonly used in ecological studies require sampling multiple transects and none of the existing protocols have taken statistical power into account.

Intensive sampling as performed by current protocols provides high precision in measures at individual transects, such as relative species encounters. High precision in response variables lead to increased power of statistical tests associating termites and the environment, such as ANOVAs and regressions due to the reduction in sample variance [5]. The most successful of all termite protocols was created by Jones and Eggleton (2000; JE) [2] based on Davies (1997) [6] and Eggleton et al. (1997) [7] and consists of surveying 20 contiguous 2 m × 5 m sections in a 100 m long transect. A quick searching survey on Google Scholar reveals more than 350 citations for the protocol.[1] The JE protocol is a rapid method for sampling termites in tropical rainforests, primarily developed to increase the number of species and trophic groups captured using from one to three transects. One or two JE transects usually provide a precise representation of the termite assemblage in a given area [2]. In spite of the efficiency of the method, the protocol is costly when the collection of multiple transects is necessary [e.g. 8].

In ecological studies of termites, each JE transect is usually a sampling unit and 10 or more transects are surveyed for statistical testing [8–10]. To improve cost-efficiency, some ecological studies have modified this protocol [9,11,12]. These modifications usually include reducing the number of sections surveyed per transect in order to allocate resources towards collecting more transects. However, the performance of such protocols in common statistical tests have not been

* Corresponding author. Universidade Federal de Santa Maria, Centro de Ciências Naturais e Exatas, Departamento de Ecologia e Evolução, Santa, Maria, 97105-900, RS, Brazil.
E-mail address: cristian.dambros@ufsm.br (C.S. Dambros).

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evaluated, and the best design to increase statistical power is unknown.

By simulating a reduction in sampling effort (number of transects and sections per transect surveyed), we tested how the total number of transects and sections per transect affect the detection of termite trophic groups, the number of termite species sampled, and the association of termite species composition with environmental variables (soil cations and soil clay percentage). We found that the ideal sampling design for ecological studies is achieved by sampling multiple transects with less than 20 sections each.

2. Methods

2.1. Sampling design

Data was obtained from Ref. [12] and additional samples collected from the same sampling area. Fieldwork was conducted between December 2008 and May 2009 at Ducke Reserve (RD; 02°55′ and 03°01′S, 59°53′ and 59°59′W). The reserve has topographic and edaphic variation typical of many areas in central Amazonia and has a total area of 10,000 ha situated northwest of Manaus (Fig. S1). The topography varies from 39 to 110 m. The nutrient-poor soils are classified in yellow clay latosol, located at the plateaus and clay-sand soil located on the ridges, which grades to sandy soils (hydromorphic podzol) in the valleys. The vegetation contains relatively uniform dense upland (terra-firme) rain forest [13], as it is not subjected to periodical inundations [14]. The only special type of vegetation recognized is locally called "campinarana" (low canopy forests adapted to extremely poor sand soil). The undergrowth is characterized by abundant stemless palms, such as *Astrocarium* spp. and *Attalea* spp [13]. The climate is characterized by a rainy season from November to May, with a relatively dry season (less than 100 mm of monthly rainfall) occurring from July to September [13]. Mean daily air humidity and mean daily temperature between 2008 and 2011 were 77.7% and 25.7 °C, respectively (Coordination of Environmental Dynamics, INPA).

2.2. Sampling design

Ducke Reserve contains a grid of six regularly spaced north-south and six east-west trails used by the Brazilian Biodiversity Research Program (PPBio; http://ppbio.inpa.gov.br). Each trail of the grid is 5 km-long, forming a 5 × 5 km grid. The grid allows access to 30 250-m long transects located 1 km apart along the trails. Transects follow the terrain contour to minimize variation in soil conditions within transects (RAPELD Method [15]; Fig. S1). Most edaphic conditions and biotic data do not show spatial autocorrelation in the area with this spatial arrangement of transects. Other PPBio researchers have made biotic and abiotic data available, allowing the evaluation of the environmental variables affecting the termite community.

Termite data from the sampling grid was taken from Ref. [12], which evaluated the effect of edaphic variables on termite diversity. In each of the 30 permanent plots, termite sampling was conducted in a 250 m long and 2 m wide transect divided into 10 regularly spaced sections of 5 m × 2 m (hereafter called T1; Fig. 1; modified from Ref. [2]). Using this method, we quantified the number of termite species represented in trophic groups (see description below), estimated termite species richness, and evaluated the association of termite community composition with soil cations and soil clay content. We then tested if a less intensive sampling effort would be efficient to estimate the measures of termite diversity captured with the maximum effort (300 sections in 30 transects; see Sampling rarefaction section). In all scenarios investigated using T1, sections within a transect were non-contiguous and separated by at least 22.2 m from one another. Transects were also maintained at least 1 km apart from each other. The minimum distance between transects ensured the independence of transects both in predictor and response variables (see Analysis section).

In addition to T1, we sampled termites using one transect as proposed by Jones and Eggleton [2] (hereafter called JE). For JE, 20 contiguous 2 m × 5 m sections were surveyed in a 100 m long transect close to the center of the sampling grid (Fig. 1). The JE protocol was sampled concomitantly to T1 transects in December 2008.

In both protocols, each section was surveyed by actively searching by three trained people for 20 min, resulting in 1 person-hour of sampling per section. Several microhabitats were searched: litter, humus, soil materials, branches and twigs, dead logs, and arboreal and subterranean nests. Nests above 2 m above ground level were not surveyed. Two sampling periods were used: wet-season in December 2008, and dry season in May 2009. Termites were manually sampled and placed in vials filled with 70% EtOH.

Individuals were sorted to genus using [16,17] and then to species or morphospecies after comparison with the collections at the Universidade Federal do Rio Grande do Norte, UFRN, and Instituto Nacional de Pesquisas da Amazônia, INPA, Brazil. Non-described species were first identified to genera level. A unique identification for each morphospecies was provided based on differences in morphological characters among species. For the Apicotermitinae subfamily, worker guts were dissected and identification was based both on external and internal morphology following the distinctive characters proposed by Ref. [18]. Apicotermitinae species were not included in the analysis of trophic groups due to the limited information in species biology.

Species were classified into trophic groups following [7,19]. For individual species and genera not included in Refs. [7,19], the classification was based on personal observations of nesting habits and workers' mandible morphology, on species descriptions, and in studies that described termite foraging and nesting habits [8,16,20]. Four trophic groups were recognized: 1) wood feeders (feeding on moderately decayed wood); 2) decayed wood feeders (feeding on decayed wood; interface soil/wood in Ref. [2]); 3) humus feeders (feeding on organic material contained in the humus; soil-feeders in Ref. [2]); and 4) leaf-litter feeders (consuming dead leaf and only occasionally green vegetation, represented by *Syntermes* spp. and *Velocitermes* spp.). Termite data are available as supporting information (Table S1).

Soil cations (K, Ca, and Mg [mg/dm^3]) and soil clay percentage were measured in previous studies, and their influence on termite species composition was tested. To obtain these environmental data, six superficial soil samples were obtained along the dentral line of each transect. The samples were obtained to a depth of 5 cm after the removal of roots and litter and then combined for each transect. The combined soil in each transect was sieved (2 mm mesh), dried at 105 °C, and analyzed at the Soil Laboratory of the Agronomy Department at INPA. The methodology for soil clay content (%) is described in Ref. [21], and soil nutrients in Ref. [22]. These data are available at https://ndownloader.figshare.com/files/3299696 and http://ppbio.inpa.gov.br.

2.3. Analysis

The performance of a sampling method was accessed by its ability to provide unbiased estimates of common parameters of termite community. For each method, we measured (1) the total number and the proportion of encountered termite species relative to the total expected for the area; and (2) the representation of trophic groups of termites. For the T1 protocol, we also quantified (3) the number of transects and sections per transect necessary to detect an association of soil clay percentage and soil cations with termite species composition. Because the third aspect requires multiple samples using the same protocol, it was not tested using the JE protocol. We used one JE transect (20 sections) and two T1 transects (10 sections per transect) for quantitative comparisons of species richness and representation in trophic groups between the two protocols.

The expected number of species for the area was calculated using the non-parametric sample-based richness estimator Chao 1 [23]. To

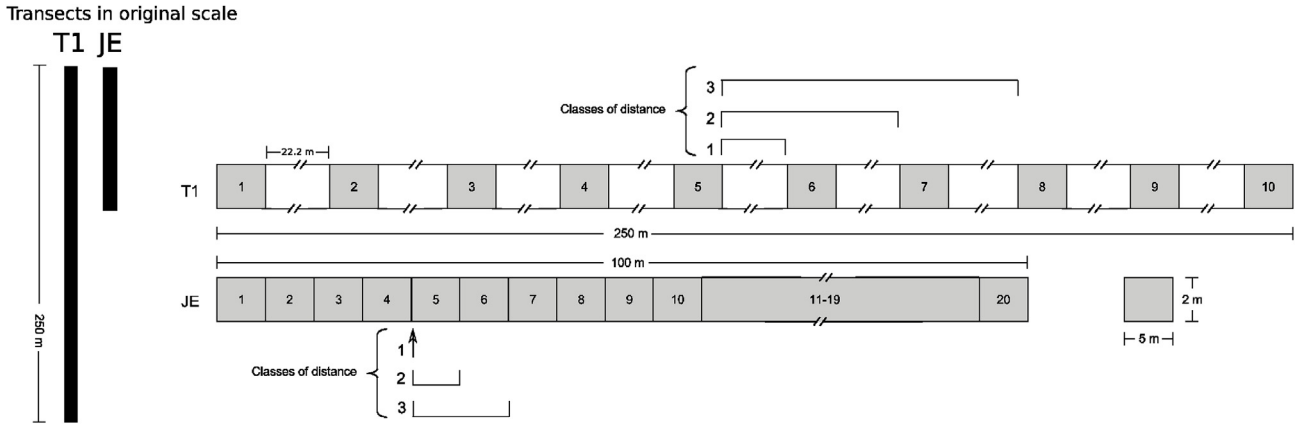

Fig. 1. Sampling protocols showing transects and sections surveyed. Distance classes ranging from 1 to 29 were used in mantel tests to test for spatial autocorrelation in species composition between sections within transects. Distance classes differ in absolute distance values in T1 and JE.

quantify termite species composition, we calculated the Jaccard similarity index between all pairs of transects (870 pairs) and then summarized the information using a Principal Coordinates Analysis (PCoA). We used the scores of the first axis of PCoA as the response variable in regression models. The Jaccard index is strongly affected by the number of rare species [24], which are difficult to detect when sampling effort is low. Therefore, the Jaccard index should be more affected by the reduction in the number of sections than most other pairwise indices of species similarity available.

The environmental gradients of soil cations (mg/dm^3 of K, Ca, and Mg summed) and soil clay percentage were used as predictor variables in simple regression models. Soil cations are weakly associated with changes in termite species composition in the area, whereas soil clay percentage is one of the variables most strongly associated with termite species composition [12]. Because *p-values* are lower for variables with stronger association with termite species composition (i.e. effect size [5]), we expect the reduction in sampling effort to affect more strongly the association of termite species composition with soil cations than with soil clay percentage.

2.4. Sampling rarefaction

To evaluate how the reduction in the number of sections and transects affect the characterization of species in trophic groups and the power of statistical tests aiming to associate termite species composition with environmental variables, we rarefied termite data by randomly reducing the number sections per transects and the number of transects (i.e. simulated scenarios with lower sampling effort). Rarefaction is a procedure of removing samples or individuals from the total pool of samples and is frequently used to evaluate how differences in sampling effort affect ecological parameters [25].

In 30 transects with 10 sections each, there are 300 possible scenarios that can be produced by rarefying transects and sections while maintaining all transects with the same number of sections. For each scenario, we randomly sampled a given number of transects (N = {1, 2, …, 30}) and sections (K = {1, 2, …, 10}). The randomization procedure was repeated 999 times for each of the 300 scenarios (e.g. 30 transects with 1 section each) for a total of 299,700 repetitions. In each repetition, the number of species in trophic groups and the association of species with the environment were recalculated.

By randomly selecting transects and sections, we obtained a null distribution for the number of species expected in each trophic group for a given scenario (i.e. combination of number of transects and number of sections per transect). To determine whether the JE protocol capture a different proportion of species in trophic groups than T1, we compared the number of species found in each trophic group using the JE protocol to the expected number of species obtained using two T1 transects (total of 20 sections). For each trophic group, *p-values* were calculated as the probability of finding the observed or more extreme differences in the number of species between the JE and two T1 transects, given that no difference exists (null hypothesis):

$$Pvalue_t = \frac{(\sum_{i=1}^{r} (ST1_{ti} - ST1_t) \geq (SJE_t - ST1_t)) + 1}{r + 1} \quad (1)$$

where SJE_t and $ST1_t$ represent the number of species in trophic group t using the JE and two T1 transects and i represents the repetition ranging from 1 to 999 (r = number of repetitions).

In each repetition, we also calculated the Jaccard similarity index between all pairs of plots and summarized the pairwise similarity index using a PCoA. The first PCoA axis was then regressed against soil cations and soil clay percentage, and the *p-values* from these associations recorded. The frequency in which a *p-value* below the usual 0.05 cut-off was obtained was used as a measure of relative statistical power, that is, the probability of rejecting a false null hypothesis. Note that actual statistical power would only be obtained using simulations because the alternative hypothesis is known a priori to be true. The method used in this study would produce qualitatively the same results if the simulated data had a similar structure of the observed data (eg. same variance in the data, same effect size, etc.).

Adjacent sections within a transect were independent in species composition from each other (Fig. S2), and rarefying non-contiguous sections in a 100 m long transect produced identical results to rarefying non-contiguous sections in a 250 m long transect (results not shown). Therefore, rarefaction was performed without taking transect length into account, similar to most other studies using richness estimators and rarefactions which also assume independence of sections within transects (eg. Ref. [2]).

All analyses were performed in the R program [26] using the Vegan package [27].

3. Results

A total of 80 termite species were collected from all sampling at the site (JE and T1 protocols). The estimated number of species using the Chao index was 104. Using two 250 m-long transects with 10 sections of 5 m × 2 m each (T1), we captured in average 26% (S = 21) of the total number of termite species found in the entire surveyed area using the maximum effort (30 transects and 300 sections), and 20% of the total estimated for the area using the sample-based Chao index. With the JE protocol, we captured 24% (S = 19) of the total number of species found using all transects combined and 18% of the estimated number of species for the area.

Species accumulation curves did not reach a plateau in any protocol when rarefying the number of sections in transects (Fig. 2). The

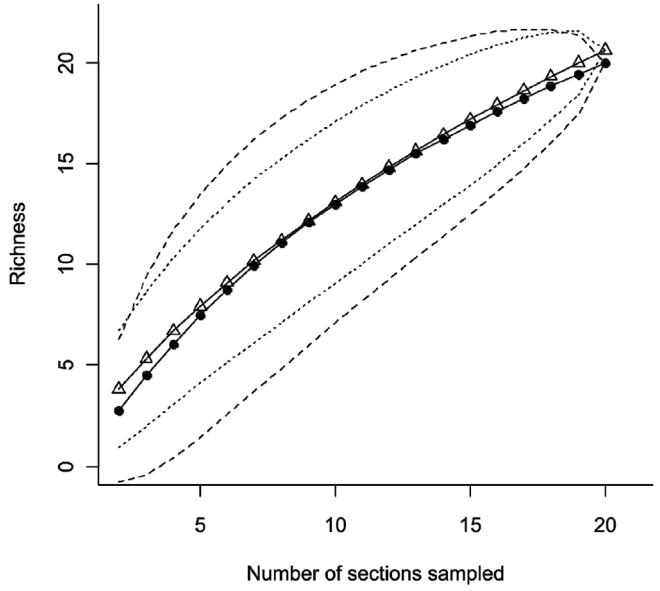

Fig. 2. Species accumulation curves comparing the sampling of 20 sections in one plot using the JE protocol (filled circles; dashed line), or 20 sections spread in two transcects using the T1 protocol (open triangles; dotted line).

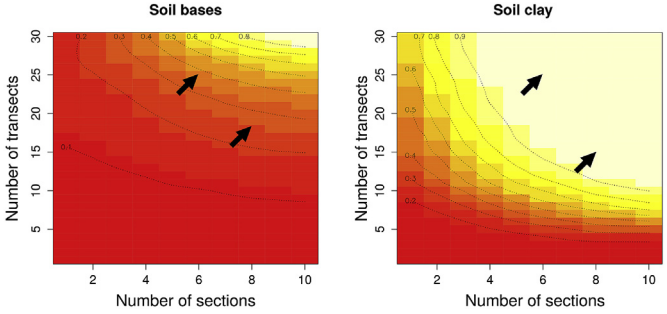

Fig. 4. Effect of the number of transects and sections surveyed on the power to detect an association of soil cations (K, Ca, and Mg) and soil clay percentage with termite species composition. Termite species composition was summarized as the first PCoA axis calculated using the Jaccard pairwise similarity index between all pairs of transects. Power was measured as the frequency in which a p-value lower than 0.05 was obtained when rarefying transects and sections. The upper arrows represent the sampling of 25 transects with six sections each (total of 150 sections surveyed). The lower arrows represent the sampling of 18 transects with eight sections each (total of 144 sections surveyed). These two combinations of sampling designs (more transects vs more sections) indicate two scenarios in which the number of transects and sections per transect would require similar sampling effort. Lighter colors represent higher statistical power. Investing sampling effort towards sampling more transects instead of sampling more sections usually increases statistical power (upper arrow in lighter colors). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

maximum number of species found using 30 T1 transects corresponded to 76% of the total species estimated by the Chao index for the entire area using all available data. With fewer than 10 sections per transect, the number of species found was reduced, but it was still possible to capture 29% of the total number of estimated species by sampling 30 transects with one section each. In general, spreading 20 sections in multiple transects slightly increases the number of species found (Fig. 2; Fig. S3).

Using the JE protocol, wood-feeding termites were the group with the highest number of species, followed by decayed-wood, humus, and leaf-litter-feeders (Fig. 3). In T1, the distribution of wood and leaf-litter feeders was similar to JE, but decayed-wood and humus feeders were more common in T1 than in JE transect (Fig. 3). When two transects with 10 sections each are sampled using T1, the number of sampled species and the distribution of trophic groups is similar to the distribution from JE (Fig. 3; $p > 0.52$ for all trophic groups), suggesting that differences between protocols could be attributed to sampling effort (i.e. more sections sampled provides a better representation regardless of sampling method; Fig. S4). Although the complete sampling of humus feeders require more sampling effort than other groups, all groups are better represented with the increase in number of sections or transects surveyed (Figs. S4–S5).

3.1. Species-environment relationship

The sampling effort necessary to detect the association of environmental variables with termite species composition, as measured by the first PCoA axis, varied depending on how strongly the environmental variable was associated with termite species composition. The association with soil clay percentage would be detected 90% of the time when sampling 30 transects with 5 sections each (Fig. 4, right). In contrast, the weaker significant association of termite species

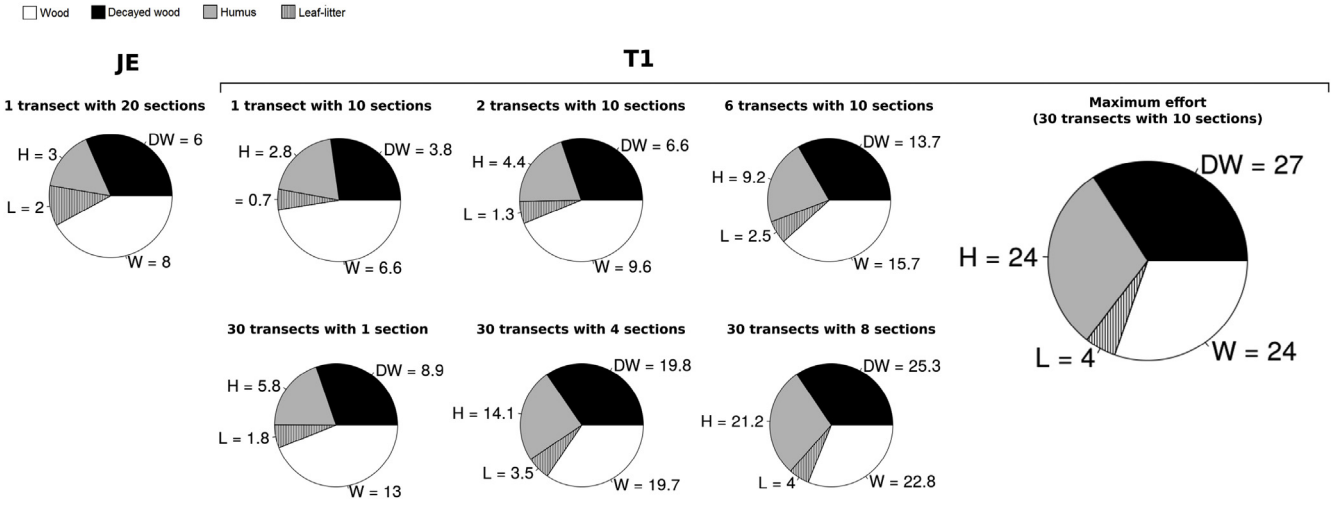

Fig. 3. Percentage of termite species in trophic groups when sampling termites using the JE and T1 protocols. For T1, the number of sections within each transect was rarefied by randomly removing sections from transects, and transects, as indicated. Rarefaction was repeated 1000 times, and the number of species in each trophic group counted. Graphs show the mean proportion of species in trophic groups in the 1000 rarefactions. W = wood feeders; DW = decayed wood feeders; H = humus feeders; L = leaf-liter feeders.

composition with soil cations would be detected 50% of the time when sampling 30 transects with 5 sections each (Fig. 4, left). For both variables, it was almost impossible to detect any association when sampling fewer than 15 transects, regardless of the number of sections sampled per transect.

4. Discussion

Our results suggest that the number of species and trophic groups sampled is similar when using a single JE transect or when sampling 20 sections distributed into several transects. However, the results indicate that a total of 20 sections, insingle or multiple transects, is not sufficient to properly characterize termite communities or to associate measures of termite community (e.g. species composition) with environmental variables. There are small differences in the number of species found and in the distribution of termite species in trophic groups when sampling one transect with 20 contiguous sections (JE; S = 19) or two transects with 10 sections each (T1; S = 21). Nevertheless, more than two transects might be necessary to properly characterize termite assemblages in termite studies using any protocol, as evidenced by the increase of species and the change in the distribution of trophic groups found using 30 T1 transects. Only after sampling 11 or more T1 transects, the distribution of species in trophic groups approached the overall distribution obtained with maximum sampling effort (300 sections in 30 transects, Fig. S5). Although we were not able to evaluate how increasing the number of JE transects might change the distribution of trophic groups, JE and T1 protocols are likely to show similar patterns.

Similar to our results, Jones and Eggleton (2000) [2] found that the proportion of old world termite groups represented in samples change with sampling effort. However, humus-feeders were the most common group in old world termites, whereas we found most species from wood- and decayed-wood feeding groups when sampling 20 sections. The proportion of species in each trophic group differs among continents, and among regions within South America [28,29], and these differences are unlikely to result only from differences in sampling. The JE protocol was developed to avoid biases on the distribution of trophic groups [30]. However, humus and soil-feeders are rarer and more patchily distributed in central Amazonia than in the forests of Cameroon, where the JE protocol was initially tested. It is likely that the detection and proper characterization of rare groups require extra sampling effort.

Although the number of termite species found in a transect increases by sampling additional sections (Fig. 2), adjacent sections within a transect are more likely to share the same species than disjunct sections (Fig. S2 [9]). Therefore, the sampling of disjunct sections is likely to result in a higher number of species found. The increase in the number of species should occur both when sections are farther apart within a transect or in multiple transects. When sampling effort is standardized, the total number of species found in a region (gamma diversity) depends on the number of species present at individual transects (alpha diversity), and on differences in species composition among transects [31–33]. Consequently, sampling additional transects should lead to an increase in the number of species found, even when the overall sampling effort is similar [9]. Because differences in species composition are usually associated with environmental conditions [8,9,12,29], the difference in results between separating the sections of a transect and making it contiguous is more likely to be pronounced when environmental conditions differ between transects (as in our study), and become less pronounced when environmental variation within a transect is high (e.g. sections placed along elevation gradient).

4.1. Species-environment relationship

Our results suggest that statistical power depends on the interaction between the number of transects and sections per transect (curved lines in Fig. 4). The number of sections sampled per transect increased statistical power when many transects are sampled, but power is always low when few transects are sampled. We found qualitatively similar results for environmental factors with high (clay) and low (cations) variability in the area.

Our results suggest that the association of termite species composition with environmental variables is easier to detect when additional transects are sampled and that the sampling of additional transects has a stronger impact on statistical power than the sampling of additional sections within the transects. If the researcher intends to sample multiple termite species in a single intensive survey for taxonomic purposes (eg. Ref. [34]), then the JE protocol is convenient because sampling does not involve costly displacement of equipment and personnel between areas. However, ecological studies investigating the association of termite diversity with environmental variables [8,10,12,12,35] require sampling multiple transects.

Ecological studies of termites usually intend to compare termite diversity among habitats, or to associate termite diversity with environmental variables by using parametric statistical tests based on *p-values,* such as ANOVAs [10,36] and regressions [4,11]. Considering a transect as the sampling unit, at least 10 independent transects per category of the predictor variable are suggested for ANOVAs and regressions [25]. In spite of these statistical constraints, most studies of termites sample from five to 15 transects (eg. Refs. [8–10,35]).

When a low number of transects is sampled, statistical tests might fail to detect any significant association of biotic or abiotic factors with termite diversity, even when the number of species in each transect is measured with high precision and the environmental variable has a strong impact on termite diversity. For example, Neoh et al. (2015) [36] sampled termites in 12 transects in Vietnam using the JE protocol. The authors characterized termite assemblages with high precision, and show strong differences in the number of species among several forest habitats. However, no significant association between termite species richness and abundance was detected ($p > 0.05$), even though abundance explained 69% of the variation in termite species richness [36]. In addition to sample few transects with high intensity, termite studies frequently group data from multiple transects to characterize precisely local assemblages in a sampling unit (eg. Refs. [11,36]). Local measures of species richness and composition will be more precise when data is combined, but statistical power is reduced if samples are grouped and few sampling units are used in statistical analyses (Fig. 4).

In classical statistical tests, the ability to detect an association between predictor and response variables increases with the number of transects (sample size) and the magnitude of the effect being investigated (e.g. difference between habitats), and decreases as measured parameters become imprecise (e.g. high unexplained variation among transects within forest habitats [5]). When intending to detect weak associations of termite diversity and predictor variables, termite studies should maximize both the number of transects and sections sampled to increase the number of sampling units and the measurement precision per unit. However, if local measures have low variability or are not strongly affected by sampling, such as the incidence of common species and most community metrics, the number of transects sampled will determine statistical power more strongly than the number of sections per transect.

4.2. Caveats

Although the number of transects sampled in our study increased the power to detect an association between termite species composition and soil variables, sampling a high number of sections per transect might be required depending on the study objective. For instance, when trying to associate the incidence of an individual termite species with environmental gradients, it might not be possible to conduct appropriate statistical tests if the species is rare and only found in one or few transects. Moreover, precision in measures of species diversity and composition at individual transects will depend on the metric used

[24]. Metrics that depend heavily on the detection of rare species will require sampling of a high number of sections to be precisely estimated. The scope of this study is not to describe all possible analyses that can be conducted in ecological studies, and we only explored the most common metrics.

Using the Jaccard index of species similarity, we found that the number of transects sampled has a strong impact on statistical power. Other metrics of species composition are likely to be less affected by the number of sections sampled per transect. Therefore, termite studies must consider the study objectives to determine the most appropriate sampling design. Measures that rely on species counts, such as species richness and the Jaccard and Sorensen indices of species similarity, are strongly affected by detection of rare species [24,32]. Dambros et al. (2016) [12] found termite species density to be associated with the abundance of predatory ants. This association was strong and easily detected, but other weaker associations were not found [12]. In contrast to indices based on species counts, indices based on the probability of sampling individual species, such as the Simpson's diversity index, the associated Morisita-Horn index of overlap, and the Bray-Curtis index of species similarity, will be more affected by the occurrence of common species [24,32], which are easily detected even when sampling few sections per transect.

Finally, our study focused on the sampling design that maximizes statistical power without considering the costs involved in field surveys. When sampling is conducted along roads or easily accessible trails, as in our study, the spread of sections in one or more transects requires a similar amount of resources and time. However, field effort in areas of difficult access may increase dramatically when multiple transects are surveyed. Future studies must consider the design that increases statistical power within the limits of resources and time available.

The JE protocol have been extensively and successfully used in many areas of the world. The protocol has standardized the sampling procedures, allowing the comparison of studies conducted by distinct researchers. Although we advocate that statistical power in an individual study can be increased by modifying this protocol, the JE protocol will continue to be a rapid and easy method to estimate several aspects of termite diversity.

5. Conclusion

Ecological studies of termites might benefit from increasing the number of transects sampled in an area while reducing sampling effort per transect. Collectively, our results and results from other studies suggest that multiple transects are usually required to properly characterize an assemblage, either considering the distribution of species in trophic groups and the number of species sampled [2,9]. However, intensively sampling of multiple transects can be costly and time-consuming. Although we acknowledge that reducing the number of sections per transect reduces standardization among studies [37], the allocation of sampling effort towards sampling more transects increases statistical power when characterizing the association of species diversity with environmental gradients. The primary purpose of an ecological study is to answer the ecological questions proposed. The best sampling design is the one that provides the highest statistical power to answer those questions. Future termite studies aiming to associate termite communities with environmental gradients should sample at least 15 transects with at least five sections each to properly characterize the diversity x environment relationships.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejsobi.2019.103145.

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